Journal of Medical Virology 61:44–51 (2000)

Hepatitis C Virus Infection in French Hemodialysis Units: A Multicenter Study

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The aims of the study were: (i) to evaluate the prevalence of hepatitis C virus (HCV) antibodies (third generation tests) and RNA (standardized ultrasensitive RT-PCR assay) in a large cohort of hemodialysis patients, and (ii) to correlate HCV markers with bioclinical features and alanine-aminotransferase (ALT) activity. Antibodies were assayed by two methods in 1,323 patients (60% men, median age 65 years, median hemodialysis duration 3 years) attending 25 French hemodialysis centers including 9 self-care units. RNA was assayed using the Cobas Amplicor 2.0 method in pooled samples from 10 anti-HCV(–/–) patients and on individual samples from the other patients. Of the 16.3% patients (range 0–44%) tested (+/+) for HCV antibodies (anti-HCV), 2.3% tested (+/–) and 81.4% tested (–/–). 70% of the anti-HCV(+/+) patients and 3% of the HCV(+/–) patients were RNA(+). Pooled analysis revealed that 5/1077 anti-HCV(–/–) patients (0.5%) were RNA(+); all 5 displayed subsequently an increase in ALT and became anti-HCV(+/+). Mean ALT was higher (multiple of normal) in anti-HCV(+/+) RNA(+) patients than in anti-HCV(+/+) RNA(–) patients (0.46 ± 0.08 vs. 0.22 ± 0.07, $P < 0.0001$) and similar in all the RNA(–) patients, whatever their HCV antibody status. Multivariate analysis demonstrated that HCV status was linked to hemodialysis duration, previous kidney transplantation and positive anti-HBc. To summarize, the determination of the RNA status of anti-HCV(+/–) patients may have clinical relevance if a policy of isolation is contemplated. Standardized ultrasensitive RT-PCR assay combined with a pooling strategy is a promising method for use in epidemiological studies. ***J. Med. Virol. 61:44–51, 2000.***

KEY WORDS: hepatitis C virus; hemodialysis; ALT; PCR; RNA; pooling; multicenter

INTRODUCTION

A high prevalence of hepatitis C virus (HCV) infection has been reported in hemodialysis patients [Pereira and Levey, 1997]. A history of blood transfusion and the duration of hemodialysis are the main risk factors associated with HCV infection in these patients [Simon et al., 1994; Pereira and Levey, 1997]. Despite the systematic screening of blood donors for HCV antibodies and the reduced use of blood transfusion, however, de novo infections still occur. Several molecular studies have suggested strongly that patient to patient transmission occurs within hemodialysis units [Allander et al., 1994; Sampietro et al., 1995; De Lamballerie et al., 1996; Stuyver et al., 1996; Olmer et al., 1997; Le Pogam et al., 1998; Katsoulidou et al., 1999; Izopet et al., 1999]. In the general population, persistent HCV infection leads to chronic hepatitis in about 80% of infected patients, often accompanied by an increase in serum transaminase levels [Tong et al., 1995]. In hemodialysis patients, the serum alanine amino transferase (ALT) level is markedly lower than in the general population, even in patients with chronic HCV infection, [Guh et al., 1995; Yasuda et al., 1995]. This means that a single ALT activity value cannot be used as a marker in screening for HCV infection. It has been reported that 0.2–28% of hemodialysis patients may carry the virus in their blood, despite testing negative for HCV antibodies [Bukh et al., 1993; Sakamoto et al., 1993; Pujol et al., 1996; Caramelo et al., 1996; Pereira and Levey, 1997; Schneeberger et al., 1998]. Discrepancies between these studies may be attributable to technical aspects or to specific clusters of de novo infections. More sensitive and specific new virological methods have been developed recently [Fabrizi and Locatelli, 1999; Doglio et al., 1999]. Effective exposure markers (third generation HCV antibodies), and

This work was carried out on behalf of the Multicenter "VHC Hémodialyse" Group.

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Accepted 5 October 1999

markers of infectivity (a standardized and ultrasensitive assay of HCV RNA using the reverse transcriptase polymerase chain reaction [RT-PCR]) are now available.

A large multicenter study was carried out involving 1,323 patients in 25 hemodialysis units. The aim was to determine in this large group of patients: (i) the prevalence of HCV antibodies, using third generation assays; (ii) the prevalence of HCV RNA, using a standardized, semi-automated, ultrasensitive RT-PCR plus a pooling strategy; and (iii) the correlation between viral parameters and ALT levels and other clinical data.

PATIENTS AND METHODS

Study Design

A total of 1323 patients on chronic hemodialysis were enrolled in 25 French hemodialysis units. All patients who met the following inclusion criteria were included: over 18 years of age, having given informed consent, undergoing regular hemodialysis for terminal chronic renal failure. The patients recruited were attending hemodialysis centers (78%) or self-care satellite units (22%). Patients on home hemodialysis or undergoing chronic peritoneal dialysis were excluded. On the day of inclusion, blood was collected before the hemodialysis session and the serum was separated without delay. One fraction was used for transaminase determination and serum tests, and the other fraction was divided into aliquots and stored frozen at −80°C until it underwent RNA testing. Samples from patients with positive or discordant HCV serology data were individually tested for HCV RNA. Samples from patients who tested negative were tested for HCV RNA in pools from ten patients. HCV RNA was also individually assayed for all patients with elevated ALT (n + 24) and also in all the patients from 3 hemodialysis units with normal ALT (n + 164).

Bio-Clinical Data

The following data were determined: hepatitis B surface antigen (HBsAg) and hepatitis B core antibody (anti-HBc) status, HIV antibody status, number and date of blood transfusions or renal transplant, time on hemodialysis and previous alpha-interferon (αIFN) therapy for HCV infection.

Biochemical Parameters

Alanine amino transferase (ALT) was assayed in each hemodialysis unit using routine automated spectrophotometric methods. The upper reference ranges (URR) were recorded in each laboratory. The activity was expressed as a multiple of the normal value (patient ALT activity/URR ratio).

Serum Assays

HCV antibodies were tested in each laboratory using two of the third-generation assays licensed by the French Medicines' Agency: HCV EIA 3.0 (Abbott Diagnostics, Rungis, France), IMX HCV (Abbott Diagnostics, Rungis, France), Ortho HCV 3.0 ELISA (Ortho Diagnostic Systems, Roissy Charles de Gaulle, France), MONOLISA anti HCV PLUS (Sanofi Diagnostics Pasteur, Marnes la Coquette, France) and Murex anti-HCV III (Murex Diagnostics, Chatillon, France). The frequency that each test was used was 17–24%. In each case, the ratio of the optical density (OD) of patient sample/OD of cut-off value was computed. The test was considered to be negative if both ratios were <0.8, to be positive if both ratios were >1.2, and classified as discordant in other cases.

HCV RNA Detection

HCV RNA was assayed in the Virology Laboratory of Toulouse University Hospital, France. The new standardized ultrasensitive RT-PCR kit HCV test 2.0 was used on the COBAS AMPLICOR system (Roche Diagnostics, Meylan, France) [Doglio et al., 1999]. The lower limit of detection of this assay is 100 copies/ml. HCV RNA was assayed directly for the patients with positive or discordant HCV serum data, whereas the pooling strategy described below was used for those who tested negative. Two hundred μl samples from ten patients were pooled. The pooled samples were centrifuged at $24{,}000 \times g$ for 60 min at 4°C. The supernatant (1800 μl) was discarded, and the pellet was then re-suspended in the remaining 200 μl. The re-suspended pellet was then processed in the same way as the individual samples: lysis, addition of an internal quantification standard, RT-PCR, and detection of the amplified products. Pooled samples classified as testing negative if the OD for HCV was <0.2 and the OD for the internal quantification standard was ≥ 0.3. If the OD for the internal quantification standard was <0.3, or the OD for the HCV RNA was ≥ 0.2, each of the samples constituting the pool was tested individually. Serial twofold dilutions of sera with a known HCV RNA copy number showed that the lower detection limit of this protocol matches that of the Cobas Amplicor 2.0 for individual serum samples, i.e., 100 copies/ml.

Statistical Analysis

Serum transaminase values were transformed logarithmically to obtain a normal distribution, and then expressed as the geometric mean in their original units by using the antilogarithms. The relationships between qualitative variables were tested using the χ^2 test. The relationships between the quantitative variables were tested using Student's *t*-test. Logistic regression models were used to evaluate the relationship between the HCV status and bioclinical data. Factors that were found to be significantly linked to HCV status by the univariate analysis were subjected to a multivariate analysis to find out which factors independently predict HCV status. A P value less than 0.05 was taken to be statistically significant.

RESULTS

Prevalence of HCV Markers

There were 1,323 patients, 792 of whom were men (60%). The median age was 65 years (range 18–96) and

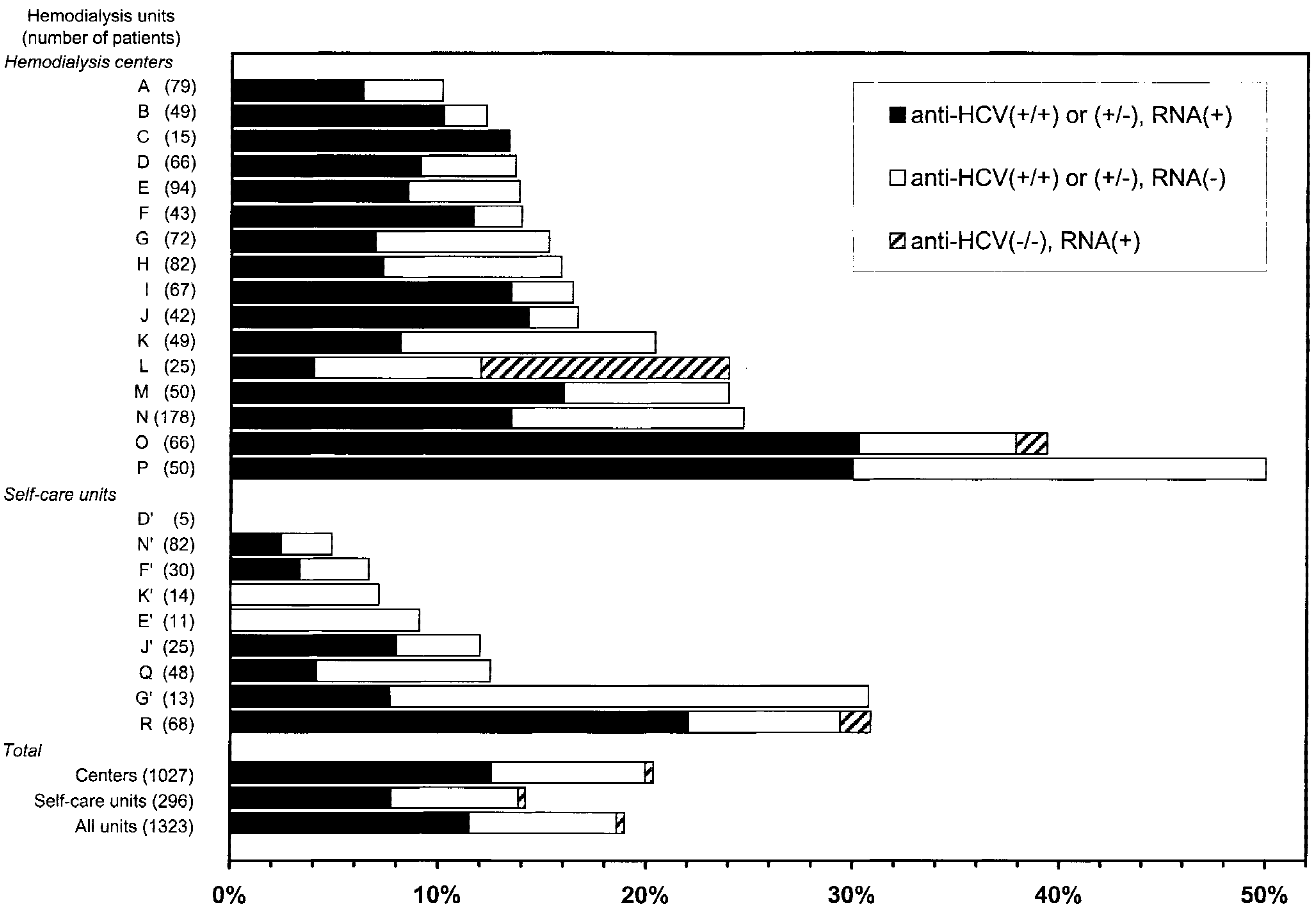

Fig. 1. Prevalence of HCV markers in 1,323 patients in HD units.

the median follow-up time of hemodialysis was 36 months (range 1–347). As shown in Figure 1, there was a high degree of scatter in HCV-marker prevalence for the various hemodialysis units. HCV antibodies were detected using two third-generation assay methods for 216 of the 1,323 hemodialysis patients (16.3%) (Table I). A positive outcome was found in both tests in 0–44% of patients. In 1,077 patients (81.4%) the HCV serum test was negative, and in the remaining 30 patients (2.3%), HCV serum test results were discordant. No difference was observed in the results obtained with the different third generation tests. No particular combination of assays favored a discordant result. HCV RNA was detectable in 151 out of the 216 (70%) patients who were positive, versus only one out of 30 patients with discordant HCV serum test data. HCV RNA was assayed in the anti-HCV(–/–) patients after pooling ten samples. Five pools out of 108 were HCV RNA(+). In each positive pool, only one serum sample was HCV RNA(+). A total of five out of 1,077 anti-HCV(–/–) patients (0.5%) tested HCV RNA positive. Two patients had above-normal ALT activity when they were included and this continued to increase during the remaining three weeks of follow-up. Four patients were experiencing an acute phase of HCV infection and seroconverted between one and four months later. The remaining patient was also infected with HIV-1 and developed anti-HCV antibodies during follow-up after starting active antiretroviral therapy. In an independent determination, HCV RNA was assayed individually in all HCV negative patients with elevated ALT values, and in HCV negative patients with normal ALT from 3 different dialysis units. HCV RNA was found to be positive in 2 out of 24 patients with elevated ALT who subsequently seroconverted, and in 1 out of

TABLE I. HCV Markers and ALT Activity in 1,323 HD Patients

Anti-HCV antibodies[b]	n (%)	HCV RNA Negative	HCV RNA Positive	ALT[a] ≤N	ALT[a] >N
Negative	1077	1072		1050	22
	(81.4)		5	3	2
Discordant	30	29		29	0
	(2.3)		1	1	0
Positive	216	65		62	3
	(16.3)		151	134	17
Total	1323	1166	157	1279	44
	(100)	(88.1)	(11.9)	(96.7)	(3.3)

[a]Number of patients with ALT activity inferior (≤N) or higher (>N) than the Upper Reference Range, on the day of inclusion.
[b]Anti-HCV antibodies were tested with two techniques, considered as positive when both were positive (patient OD/cut-off OD > 1.2), negative when both were negative (patient OD/cut-off OD < 0.8) and discordant in all the other cases.

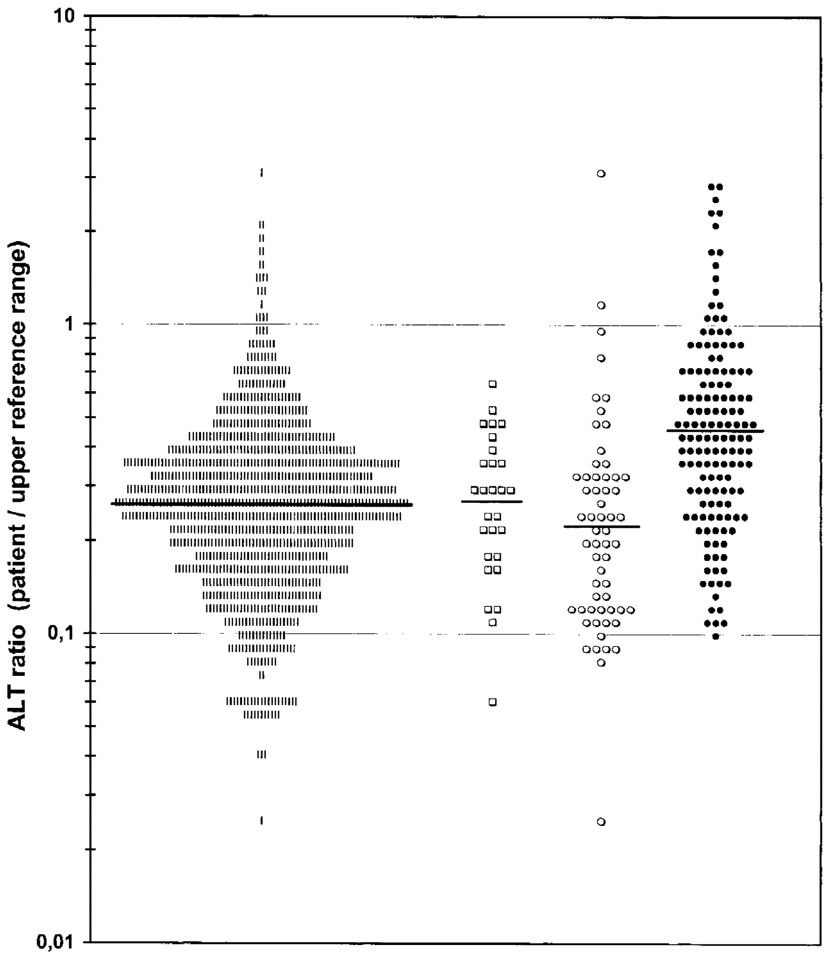

Fig. 2. Scattergram of ALT ratios in 1,272 HD patients with negative HBs Ag according to HCV status. Each symbol corresponds to one individual ration (patient ALT activity/upper reference range), collected on the day of inclusion. Bars (|) correspond to patients testing negative in serum tests, squares (□) to patients with discordant serology data and circles (○, ●) to patients testing positive. Solid circles (●) correspond to patients with detectable HCV RNA, whereas hollow symbols (□, ○) correspond to patients with no detectable HCV RNA. Patients with detectable HCV RNA and negative (n + 5) or discordant (n + 1) serum tests data were excluded from this analysis. In each group of patients, horizontal lines represent the mean ratio.

164 HCV negative patients with normal ALT. This latter patient had been infected with both HCV and HIV. Pooling and individual testing strategies gave completely consistent outcomes.

Correlation Between HCV Markers and ALT Levels

As illustrated in Figure 2, after excluding patients with a positive HBs Ag (n + 44), ALT activity was computed as a multiple of normal in the various groups of patients. The mean ALT activity (0.46 ± 0.08) in the anti-HCV(+/+) RNA(+) patients was significantly higher than in anti-HCV(+/+) RNA(–) patients ($P <$ 0.0001). Mean ALT values were not found to be significantly different in RNA(–) patients who were anti-HCV(–/–) (0.26 ± 0.01), anti-HCV(+/–) (0.28 ± 0.07) or anti-HCV(+/+) (0.22 ± 0.07); however, ALT activity was >1 in only 16 out of 149 anti-HCV(+/+) RNA(+) patients (11.2%).

Correlation Between HCV Markers and Bioclinical Data

No gender-related difference was found in the prevalence of anti-HCV antibodies , but patients with anti-HCV(+/+) or (+/–) were significantly younger than the HCV(–/–) patients (Table II). The prevalence of anti-HCV(+/+) or (+/–) was lower in self-care satellite units than in hemodialysis centers. Patients with testing HCV(+/+) or (+/–) had a higher percentage of previous kidney transplantation, had received more blood transfusions and more blood units and had a higher prevalence of anti-HBc and HBs Ag (Table II). Finally, there was a strong relationship ($P < 0.0001$) between the prevalence of anti-HCV(+/+) or (+/–) and the duration of dialysis (Fig. 3).

Factors identified by univariate analysis to be associated with anti-HCV(+/+) or (+/–) i.e., (i) dialysis duration, (ii) anti-HBc status, (iii) previous kidney transplantation, (iv) HBs Ag status, and (v) previous blood transfusions were also tested using multivariate analysis. The only factors that were still found to be significantly associated with positive or discordant serum test data were the duration of hemodialysis, the presence of anti-HBc and a previous history of kidney transplantation (Table III).

DISCUSSION

HCV infection remains a major problem in hemodialysis units. Studies intended to determine the prevalence of HCV markers have given conflicting results,

TABLE II. Correlation of Anti-HCV Antibodies With Bio-Clinical Data

	Anti-HCV antibodies			
	Negative	Discordant	Positive	P[a]
Mean age (SD)	63.0 (16)	61.7 (17)	57.9 (16)	<0.001
Dialysis in centers (%)	76.3	80.0	82.6	<0.05
Mean years of HD (SD)	3.8 (4.3)	7.0 (7.1)	12.4 (7.7)	<0.0001
Renal transplantation antecedent (%)	5.1	16.7	25.5	<0.001
Blood transfusion antecedent (%)	62.8	63.0	79.5	<0.05
Mean number of transfusions (SD)	6.8 (9.3)	9.0 (8.3)	12.3 (21)	<0.05
Positive HBs antigen (%)	2.7	3.6	6.6	<0.01
Positive anti-HBs antibodies (%)	9.4	23.3	34.9	<0.001

[a]Refers to patients with either positive or discordant serology compared to patients with negative serology.

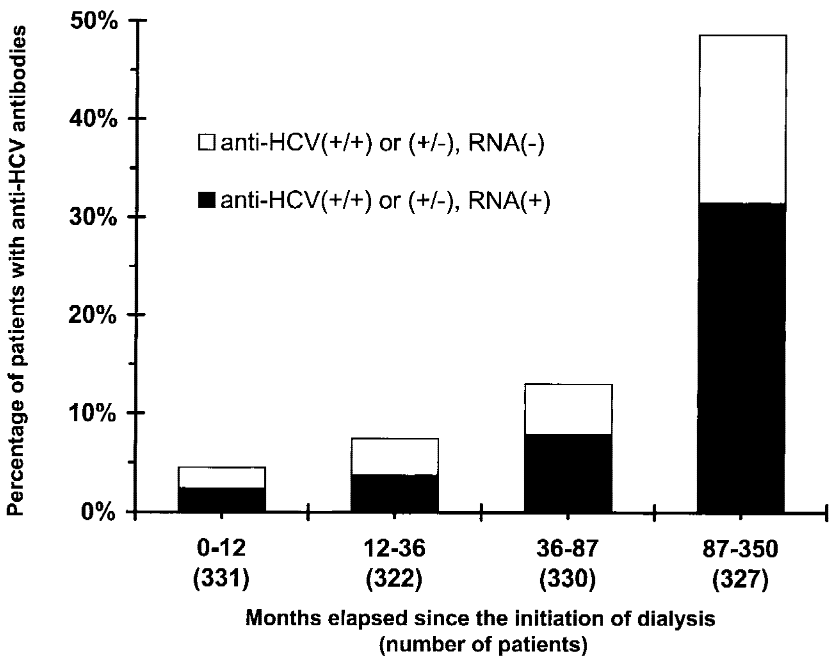

Fig. 3. Analysis of the correlation between HCV markers and the duration of dialysis. HD patients are subdivided into quartiles according to the duration of dialysis. Boxes correspond to percentage of patients with positive or discordant serum test data. Solid boxes (■) correspond to patients with detectable HCV RNA and hollow boxes (□) to patients with no detectable HCV RNA.

TABLE III. Factors Associated With Anti-HCV (+/+) or (+/–)

Variable	Odds ratio	95% Confidence interval	P
Univariate analysis			
Dialysis duration[a]	3.10	2.67–3.80	<0.0001
Renal transplantation antecedent	6.10	4.08–9.12	<0.001
Blood transfusion antecedent	1.47	1.10–1.98	<0.05
Number of blood units transfused[a]	1.74	1.35–2.23	<0.05
Positive HBs antigen	2.35	1.24–4.45	<0.01
Positive anti-HBc antibodies	4.87	3.47–6.83	<0.001
Multivariate analysis			
Dialysis duration[a]	2.51	2.09–3.02	<0.0001
Positive anti-HBs antibodies	2.62	1.76–3.91	<0.001
Renal transplantation antecedent	2.12	1.33–3.39	<0.01

[a]Calculated after logarithmic transformation.

that could reflect heterogeneity in the performance of serology and molecular tools, or peculiar epidemiological clusters. To obtain a better estimate of the prevalence of HCV infection in French hemodialysis units, a large-scale survey was conducted using recently-developed virology tools such as third generation anti-HCV antibodies, and a standardized semi-automated ultra-sensitive RT-PCR assay for HCV RNA testing.

In accordance with the French legal requirements at the beginning of the study, HCV antibodies were assayed in each sample by two methods and 16.3% of the patients tested positive by both third-generation assays. As reported previously [Pereira and Levey, 1997], however, the prevalence of HCV antibodies was not uniform in the various units in this study, and ranged from 0–44%. More than 30% of the patients who were

positive were HCV RNA(−). This figure is higher than that reported in previous studies [Pol et al., 1993; Simon et al., 1994; Pereira and Levey, 1997; Schneeberger et al., 1998]. This may be explained, however, by the effectiveness of previous αIFN therapy. The virus had been eradicated in 15 out of the 24 treated patients (62.5%) [Izopet et al., 1997]. A similarly high rate of HCV RNA clearance in hemodialysis patients has also been reported by others [Raptopoulou-Gigi et al., 1995] and is probably related to the impairment of αIFN pharmacokinetics in hemodialysis patients [Rostaing et al., 1998]. Because ALT activity remained completely normal and HCV RNA was repeatedly negative in these patients, it can be assumed that these patients had lost the HCV. Discordant data were obtained in 2.3% of the patients in the two anti-HCV assays. HCV RNA was negative in all but one of these patients. A discrepancy of this type could result from a non-specific cross-reaction in one serum test or could reflect the decrease of antibody titer after a previous infection and spontaneous clearance. Interestingly, patients with either anti-HCV(+/−) or anti-HCV(+/+) RNA(−) presented similar features with regard both to their bioclinical history and their ALT levels.

The prevalence of HCV antibodies was closely related to the time on dialysis, as reported previously [Niu et al., 1993; Dussol et al., 1995; Pujol et al., 1996; Nakayama et al., 1996; Fabrizi et al., 1997]. Most of the patients with HCV markers had probably been infected in the past, either as a result of blood transfusions before the screening tests were introduced, or by nosocomial infection. A high prevalence of HCV antibodies was observed in patients with a history of kidney transplantation. HCV infection may have been transmitted to these patients from an infected organ donor [Pereira and Levey, 1997] or, more probably, by a transfusion because blood is often required during transplantation. An association between the age of the patients and their HCV status was also found. This finding conflicts with the findings of previous studies [Nakayama et al., 1996; Fabrizi et al., 1997]. In the present study, a high proportion of patients were in the older age group (median age 65 years). Unlike younger patients, most of them are not infected with HCV, they have a short history of hemodialysis and blood transfusions are avoided by erythropoietin therapy. The correlation between HCV and HBV status has been addressed in a few studies that have given conflicting results [Guh et al., 1995; Nakayama et al., 1996; Pujol et al., 1996; Fabrizi et al., 1997]. Anti-HBc and HBsAg were positive more frequently in anti-HCV(+) patients. Because all hemodialysis patients included recently have been vaccinated against HBV infection, the association between these markers may also reflect a past exposure to both viruses before secure hemodialysis processes were introduced. Finally, patients found positive have undergone more often previous transfusions and have received more blood units. Multivariate analysis revealed that a history of transfusion was not an independent predictive factor for a positive HCV positive serum test and hemodialysis duration was a stronger predictive factor, as previously reported [Pereira and Levey, 1997]. In accordance with other reports [Jadoul et al., 1993; Simon et al., 1994; Pujol et al., 1996], half of the infected patients had no history of transfusion. This suggests strongly that the transmission of HCV is nosocomial [Allander et al., 1994; Sampietro et al., 1995; De Lamballerie et al., 1996; Stuyver et al., 1996; Olmer et al., 1997; Le Pogam et al., 1998; Izopet et al., 1999; Katsoulidou et al., 1999] and it is still a major issue in hemodialysis units.

New standardized, reproducible and ultrasensitive assays are now available to determine HCV RNA status [Doglio et al., 1999]. To the best of our knowledge, this study is the first trial of the Roche Cobas ultrasensitive assay to be conducted in a large cohort of hemodialysis patients. Moreover, a pooling strategy was used that did not lead to any loss of sensitivity, to determine the HCV RNA status of anti-HCV(−) patients. Pooling of samples before RT PCR had been described previously in hemodialysis patients in only one recent study [Schneeberger et al., 1998]. The researchers used a in-house nested PCR, however, using agarose gels to detect the PCR products. This standardized semi-automated assay would be suitable for large-scale use, especially in epidemiological studies. Furthermore, because the pooled samples were ultracentrifuged before nucleic acid extraction, pooled sera from ten patients could be analyzed with no loss of HCV RNA detection sensitivity.

Several workers have reported high proportions (7–28%) of patients who were HCV RNA(+) despite negative serum tests [Bukh et al., 1993; Sakamoto et al., 1993; Pujol et al., 1996; Caramelo et al., 1996]. These studies were all conducted, however, using second-generation anti-HCV testing or non-standardized RT-PCR procedures. In contrast, in a large recent multicenter study in which third generation anti-HCV testing was carried out, the virus was detected in the blood of only 0.23% of seronegative patients [Schneeberger et al., 1998]. In the present study, only 5 out of 1,077 patients i.e., 0.5% were anti-HCV(−)RNA(+). Four were experiencing a de novo HCV infection and seroconverted rapidly within 4 months, after an increase in ALT, and 3 of the patients were from the same unit, where nosocomial transmission is strongly suspected to occur. Severe immunodepression could account for the anti-HCV(−) status of the remaining patient, who presented with concomitant HIV plus HCV infection. After antiretroviral therapy, his immune status was partially restored, but he underwent full anti-HCV seroconversion. In the present study, therefore, a positive RNA test plus a negative HCV serum test corresponded to de novo infection rather than chronic infection.

Discrepancies have been described between the ALT level and anti-HCV antibodies [Pereira and Levey, 1997]. Baseline levels are often lower in hemodialysis patients [Guh et al., 1995; Yasuda et al., 1995; Caramelo et al., 1996; Fabrizi et al., 1997], that supports the

use of specific upper limit of normal values in hemodialysis patients [Guh et al., 1995; Yasuda et al., 1995; Fabrizi et al., 1997]. Serum ALT values were significantly higher in viremic patients, probably reflecting a link between viral replication and liver damage that has already been described [Fabrizi et al., 1997]. Only 12% of the anti-HCV(+) RNA(+) patients exhibited elevated ALT, however, when they were included. Fluctuating ALT values are common in HCV-infected hemodialysis patients [Simon et al., 1994]. Anti-HCV(+) RNA(−) patients displayed features similar to those of anti-HCV(−) patients, suggesting real clearance of HCV infection [Fabrizi et al., 1997]. These patients are repeatedly HCV RNA(−) and carry no threat of transmitting HCV to other patients, although they themselves are not protected against a new HCV infection.

To summarize, the present epidemiological survey of HCV RNA status in HCV negative hemodialysis patients showed that pooling ten sera combined with the use of an ultrasensitive, standardized RT-PCR assay provides a reliable and sensitive method. The presence of HCV RNA(+) in patients testing negative corresponded to the post-infection gap before ALT levels rose and HCV seroconversion occurred. Third generation serum assays provide a valid method for HCV testing, however, if a seronegative patient experiences any unexplained elevation of ALT above the upper limit of normal, HCV RNA status should be explored without delay. The clinical and biochemical features of the anti-HCV(+/+) RNA(−) patients differed significantly from those of anti-HCV(+/+) RNA(+) patients and were similar to those of anti-HCV(−/−) patients. This may have clinical relevance in hemodialysis units where the introduction of an isolation policy is contemplated.

ACKNOWLEDGMENTS

The authors thank all the technicians and nurses who took part in this study. We particularly thank M. Dubois for technical assistance, C. Dorbes for collecting and archiving data, and Dr. J.P. Charlet, who provided statistical advice. We are indebted to the nephrologists and biologists from the multicenter group "VHC Hémodialyse" that include: D. Verdier and C. Almayrac (Albi); F. Counillon and P. Roques (Auch); P. Donnadieu, C. Briat, P. Hue, and V. Gras (Avignon); J.M. Dueymes (Brest); R. Boudet, Y. Gontier, P. Honoré and A. Saumabère (Brive); J.C. Valery, C.P. Grasmick and C. Maheu (Cahors); G. Mestressat and J. Daleas (Lourdes); P. Giraud and P. Castelnau (Montauban); F. Dehais-Goffinet and A. Clarac (Pamiers); L. Fabre and J.M. Mansuy (Rodez); J.M. Berthonnaud and J. Journe (Tarbes); R. Ibos and P. Moinard (Toulouse Muret); F. Moussion and M. Prola (Toulouse St. Exupery-Languedoc), B. Seigneuric and J. Puel (Toulouse Purpan); J.M. Lanau (Toulouse AAIR), D. Durand, P. Bories and S. Henry (Toulouse Rangueil).

REFERENCES

Allander T, Medin C, Jacobson SH, Grillner L, Persson MAA. 1994. Hepatitis C transmission in a hemodialysis unit: molecular evidence for spread of virus among patients no sharing equipment. J Med Virol 43:415–419.

Bukh J, Wantzin P, Krogsgaard K, Knudsen F, Purcell R, Miller RH. 1993. High prevalence of hepatitis C virus (HCV) RNA in dialysis patients: failure of commercially available antibody tests to identify a significant number of patients with HCV infection. Copenhagen Dialysis HCV Study Group. J Infect Dis 168:1343–1348.

Caramelo C, Bartolome J, Albalate M, de Sequera P, Navas S, Bermejillo T, Oliva H, Marriott E, Ortiz A, Ruiz Tunon C, Casado S, Carreno V. 1 996. Undiagnosed hepatitis C virus infection in hemodialysis patients: value of HCV RNA and liver enzyme levels. Kidney Int 50:2027–2031.

De Lamballerie X, Olmer M, Bouchouareb D, Zandotti C, De Micco P. 1996. Nosocomial transmission of hepatitis C virus in hemodialysis patients. J Med Virol 49:296–302.

Doglio A, Laffont C, Caroli-Bosc F, Rochet P, Lefebvre J. 1999. Second generation of the automated Cobas Amplicor HCV assay improves sensitivity of hepatitis C virus RNA detection and yields results that are more clinically relevant. J Clin Microbiol 37:1567–1569.

Dussol B, Berthezene P, Brunet P, Berland Y. 1995. Hepatitis C virus infection among chronic dialysis patients in the southeast of France. Provence-Alpes-Cote d'Azur Nephrologists Group. Nephrol Dial Transplant 10:477–478.

Fabrizi F, Locatelli F. 1999. Hepatitis C virus in the hemodialysis units: novel insights by new techniques? Nephrol Dial Transplant 14:1072–1075.

Fabrizi F, Lunghi G, Andrulli S, Pagliari B, Mangano S, Faranna P, Pagano A, Locatelli F. 1997. Influence of hepatitis C virus (HCV) viremia upon serum aminotransferase activity in chronic dialysis patients. Nephrol Dial Transplant 12:1394–1398.

Guh J, Lai Y, Yang C, Chen S, Chuang W, Hsu T, Chen H, Chang W, Tsai J. 1995. Impact of decreased serum transaminase levels on the evaluation of viral hepatitis in hemodialysis patients. Nephron 69:459–465.

Izopet J, Rostaing L, Moussion F, Alric L, Dubois M, Ton-That H, Payen J, Duffaut M, Durand D, Suc J, Puel J. 1997. High rate of hepatitis C virus clearance in hemodialysis patients after alpha-interferon therapy. J Infect Dis 176:1614–1617.

Izopet J, Rostaing L, Pasquier C, Sandres K, Puel J. 1999. Molecular evidence for nosocomial transmission of hepatitis C virus in a french hemodialysis unit. J Med Virol 58:139–144.

Jadoul M, Cornu C, van Ypersele de Strihou C. 1993. Incidence and risk factors for hepatitis C seroconversion in hemodialysis: a prospective study. The UCL Collaborative Group. Kidney Int 44: 1322–1326.

Katsoulidou A, Paraskevis D, Kalapothaki V, Arvanitis D, Karayiannis P, Hadjiconstantiou V, Hatzakis A. 1999. Molecular epidemiology of a hepatitis C virus outbreak in a hemodialysis unit. Multicentre Hemodialysis Cohort Study on Viral Hepatitis. Nephrol Dial Transplant 14:1188–1194.

Le Pogam S, Le Chapois D, Christen R, Dubois F, Barin F, Goudeau A. 1998. Hepatitis C in a hemodialysis unit: molecular evidence for nosocomial transmission. J Clin Microbiol 36:3040–3043.

Nakayama E, Liu J, Akiba T, Marumo F, Sato C. 1996. Low prevalence of anti-hepatitis C virus antibodies in female hemodialysis patients without blood transfusion: a multicenter analysis. J Med Virol 48:284–288.

Niu M, Coleman P, Alter M. 1993. Multicenter study of hepatitis C virus infection in chronic hemodialysis patients and hemodialysis center staff members. Am J Kidney Dis 22:568–573.

Olmer M, Bouchouareb D, Zandotti C, de Micco P, de Lamballerie X. 1997. Transmission of the hepatitis C virus in an hemodialysis unit: evidence for nosocomial infection. Clin Nephrol 47:263–270.

Pereira B, Levey A. 1997. Hepatitis C virus infection in dialysis and renal transplantation. Kidney Int 51:981–999.

Pol S, Romeo R, Zins B, Driss F, Lebkiri B, Carnot F, Berthelot P, Brechot C. 1993. Hepatitis C virus RNA in anti-HCV positive hemodialyzed patients: significance and therapeutic implications. Kidney Int 44:1097–1100.

Pujol F, Ponce J, Lema M, Capriles F, Devesa M, Sirit F, Salazar M, Vasquez G, Monsalve F, Blitz-Dorfman L. 1996. High incidence of hepatitis C virus infection in hemodialysis patients in units with high prevalence. J Clin Microbiol 34:1633–1636.

Raptopoulou-Gigi M, Spaia S, Garifallos A, Xenou P, Orphanou H, Zarafidou E, Petridou P, Vrettou H, Vagionas G, Galaktidou G. 1995. Interferon-alpha 2b treatment of chronic hepatitis C in hemodialysis patients. Nephrol Dial Transplant 10:1834–1837.

Rostaing L, Chatelut E, Payen J, Izopet J, Thalamas C, Ton-That H, Pascal J, Durand D, Canal P. 1998. Pharmacokinetics of αIFN-2b in chronic hepatitis C virus patients undergoing chronic hemodialysis or with normal renal function: clinical implications. J Am Soc Nephrol 9:2344–2348.

Sakamoto N, Enomoto N, Marumo F, Sato C. 1993. Prevalence of hepatitis C virus infection among long-term hemodialysis patients: detection of hepatitis C virus RNA in plasma. J Med Virol 39:11–15.

Sampietro M, Badalamenti S, Salvadori S, Corbetta N, Graziani G, Como G, Fiorelli G, Ponticelli C. 1995. High prevalence of a rare hepatitis C virus in patients treated in the same hemodialysis unit: evidence for nosocomial transmission of HCV. Kidney Int 47:911–917.

Schneeberger P, Keur I, van der Vliet W, van Hoek K, Boswijk H, van Loon A, van Dijk W, Kauffmann R, Quint W, van Doorn L. 1998. Hepatitis C virus infections in dialysis centers in The Netherlands: a national survey by serological and molecular methods. J Clin Microbiol 36:1711–1715.

Simon N, Courouce A, Lemarrec N, Trepo C, Ducamp S. 1994. A twelve-year natural history of hepatitis C virus infection in hemodialyzed patients. Kidney Int 46:504–511.

Stuyver L, Claeys H, Wyseur A, van Arnhem W, De Beenhouwer H, Uytendaele S, Beckers J, Matthijs D, Leroux-Roels G, Maertens G, De Paepe M. 1996. Hepatitis C virus in a hemodialysis unit: molecular evidence for nosocomial transmission. Kidney Int 49:889–895.

Tong M, el-Farra N, Reikes A, Co R. 1995. Clinical outcomes after transfusion-associated hepatitis C. N Engl J Med 332:1463–1466.

Yasuda K, Okuda K, Endo N, Ishiwatari Y, Ikeda R, Hayashi H, Yokozeki K, Kobayashi S, Irie Y. 1995. Hypoaminotransferasemia in patients undergoing long-term hemodialysis: clinical and biochemical appraisal. Gastroenterology 109:1295–1300.